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Assessment of Skin Conditions Using Profilometry

by Peter L. Dorogi and Marek Zielinski
Clairol Research Laboratory, Stamford, Connecticut

From early on, scientific investigators of skin-surface topography recognized a meaningful description of surface features required specialized data reduction procedures. Indeed, much of the creative effort in this aspect of dermatological science has been on the mathematical side, not only in instrument development.

Visual examination of the skin surface reveals a system of furrows, and closer examination of the intermediate plateau regions shows the presence of secondary, finer furrow systems. Higher magnification reveals the arrangement of elementary cellular components, the corneocytes, which in turn express their own morphological features on the skin surface microtopography.

Establishment of methods to express this hierarchy of surface features in concise numerical fashion is one goal of modern profilometry. Recent developments in image analysis allow for rather sophisticated processing of either optical or electron microscopic images, and these techniques extend, in theory, to all topographical features and measurement scales. Imaging techniques, however, have an intrinsic dependence on the interaction of the illuminating beam with the surface and therefore are somewhat indirect.

Power to be objective

Established techniques of profilometry, in which the surface is measured by traversal of the surface with a stylus instrument, is relatively safe from artefactual errors. Profilometry, although not very powerful for visual demonstrations, nonetheless has the power to be objective and is easily expressible in numerical ways.

Profilometric descriptions were first developed for metallic surfaces. Adoption of these techniques to characterization of the skin surface required additional developments to satisfy new measurement and data handling needs. Two major issues confronting the profilometric technique are 1) faithful transfer of the skin surface pattern onto a harder material, and 2) identification of descriptive parameters suited for biological surfaces, which usually possess a hierarchy of spatial organization and therefore require topographical descriptors that are intrinsically independent of the scale of measurement. This problem is treated in more detail later.

Profilometric scanning of a surface usually is performed with an industrial metallurgical profilometer, such as Talysurf 4 (Rank, Taylor Hubson) or Perthometer S6P (Feinpruef). Although one relatively new type of profilometer, the Hommel Tester (Hahn and Kolb, Ltd., Rugby, UK) allows direct tracing of the skin surface with a diamond stylus of light weight, the most commonly used techniques involve preparation of a replica that then is scanned with a profilometer.

Skin replication is a two-step process, requiring prior generation of a negative impression. A study of candidate impression materials was reported a decade ago by Makki, et al.[1], who identified a silicone rubber commonly used to make dental impressions, Silflo, as being highly suitable. The procedure is described in considerable detail in an article by Cook, et al.[2] Other silicone rubbers, such as Silosoft and Xantopren also have been used with success.[3]

Positive replication of the skin's surface fea-

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tures is attained after a cast is made of the silicone rubber negative, usually with an epoxy resin or a thermoplastic polymer. Resins that have been found useful for positive casts include various Araldites[4] and Embed 812.[2,5] The group from the Department of Medicine at the University of Wales, Cardiff, where skin profilometry was pioneered, traditionally has used the styrene mounting medium DPX as the cast material.[6-10] Polyethylene also has been used, and although it is softer than epoxy resins, gives excellent rendering of skin surface details.

Like a phonograph

The actual profilometric measurement follows the same operational principles for most investigators. A transducer assembly containing the stylus is moved horizontally at constant speed along the surface of the sample, or conversely, the sample is moved under the stylus. This setup resembles a phonograph, in which a diamond-tipped stylus moves along grooves in a record, transferring the information written into the record into electrical signals. Similarly, topographical features of the surface induce vertical movements in the stylus, which then are converted into electrical signals and subsequently digitized, yielding the profile of the cross-section of the surface.

In most commercial profilometers, the rate of scanning (millimeters/second) determines the horizontal resolution (number of data points collected/millimeter, which can be as high as 10,000). In addition, the slower the scan speed the less likely it is the stylus will skip over small furrows, and the possibility for ploughing the stylus through finer surface features may also be reduced. Visual evidence of ploughing through a polyethylene replica is shown in Figure 1.

Other than selecting the proper instrumental settings, a major challenge in analyzing profilometric data has been the identification of useful data filtering techniques, with flexibility to adjust for different scales of spatial resolution.

Interpretation of data

Evaluation of skin care products in consumer use tests involves sensory awareness of various physical properties, both of the product and of the skin. The skin may appear smooth or rough, soft or stiff; it may feel too dry or flaky. The judgement of product application esthetics may, for example, sense the friction between two skin surfaces, yielding the impression of greasiness. The improved appearance of treated skin may reflect not only the topographical changes in the skin surface, but also the physical properties and distribution of product residue on the surface.

If the skin surface is covered by a product (e.g. cream), the silicone replica will not yield a faithful image of the skin surface. For this reason, profilometric measurements may not be the best suited near the time a product has been applied. Other types of measurements, such as those of skin surface friction, discussed in detail by Wolfram[11] and Gerrard,[12] may be more meaningful. For a visual analysis, imaging techniques involving microphotography, video recording and microdensitometry have become available.[10,13]

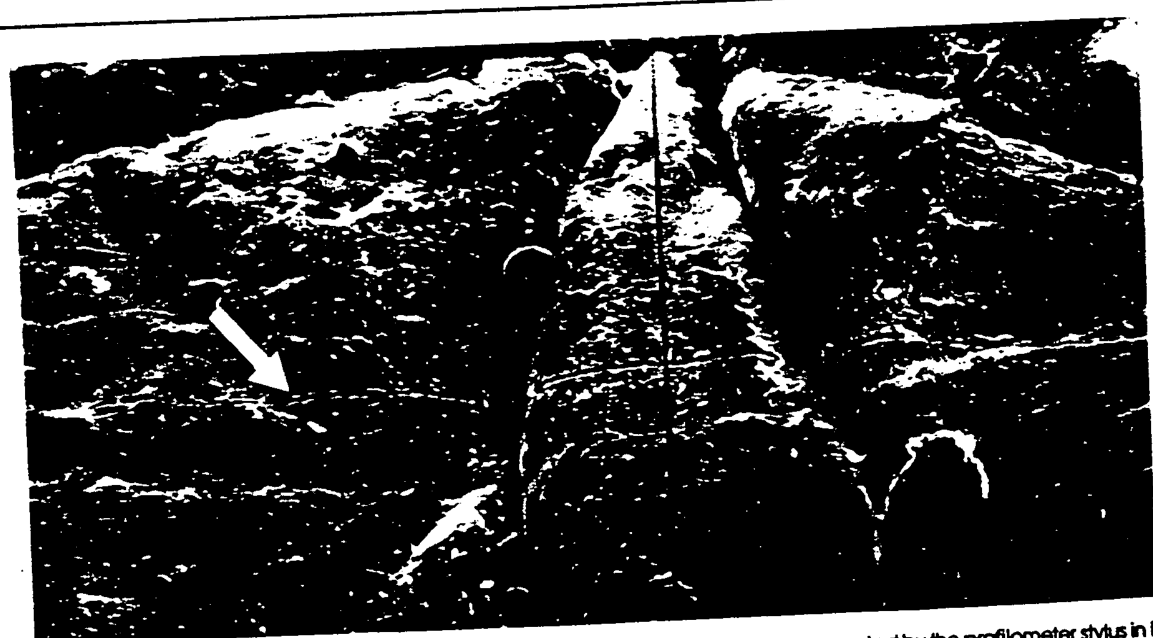

Figure 1: Electron micrograph of a replica of the skin surface. Arrow points to a furrow created by the profilometer stylus in its traversal of the replica (Microscopy by A. Hambidge, Clairol, Inc.).

The increasing acceptance of Kligman's Regression Test[14] as the standard clinical technique by which moisturizer efficacy should be assessed brings with it the opportunity to use profilometry in support of clinical data. In Kligman's method, product efficacy is judged in terms of its longevity (where the effects of treatment are monitored in time after cessation of product application), including improvements in surface texture and reduction of surface roughness.

Because the clinical assessments must be carried out at least six hours, and even days, after product has been applied, silicone rubber impressions may faithfully reproduce the topographical features of interest. The challenge then becomes the reduction of the profile to roughness parameters that can be compared and related to clinical scores.

Characterizing the surface

In profilometry, the surface of the skin is represented by a profile line, usually with highly irregular features that correspond to the peaks, valleys and plateaus encountered by the stylus in its traversal of the skin surface. As alluded to earlier, this profile contains contributions from a hierarchy of surface features, related to the multiscale morphology of the epidermis and dermis.

On the largest scale, we can see folds and wrinkles of the kind a palm reader may use to tell your future. Magnifying the image slightly, one observes a finer pattern of grooves, such as those used for fingerprinting, and a crossing network of furrows. Further magnification reveals the fine structure of the stratum corneum, which may be either smooth or scaled.

Such as analysis of surface features can be called "descriptive topography." The analyses may look at scaling disorders of the skin surface, others may evaluate the effects of moisturizers, and still others may investigate the effects of aging on the surface contour. For the most part, the fine, small-scale differences that have to be resolved for studies of scaling disorders appear to be better resolved with image analysis techniques.[10] The influence of age, on the other hand, has been studied extensively with profilometry,[2,18] and a variety of age-dependent changes can be statistically isolated from the skin relief.

Overall, the studies point to the increasing number, depth and prominence of wrinkle lines, and the disappearance of the finer surface furrows that are present in youth. The furrows appear overall narrower in older skin, and the intersection of furrows takes place at less of an acute angle, again suggesting that only the major furrow lines remain.

To quantify data

The main point in doing the profilometric measurements is, however, quantification of the data. After all, one can perform the descriptive topography simply on microscopic skin images.

Quantification of topographical changes following hydration of the skin has been reported extensively.[2,16,19,20] Most investigations confirm the initial finding by Cook[16] that dry skin is characterized by fewer peaks in the profile line than moist skin, but that the proportion of larger peaks is greater in dry skin. As noted for the effects of aging, some of the finer surface pattern also is not evident in dry skin. Superimposed on the peak-valley pattern are differences on a smaller scale, the microrelief, which reflects the larger desquamating flakes of dry skin.

It has been pointed out,[4,19] that the roughness parameters widely used to describe skin profiles fall into three categories: 1) amplitude parameters that refer to vertical aspects of the profile, e.g. peak heights and furrow depths, 2) spacing parameters that refer to horizontal distance between features along the plane of the skin surface, and 3) so-called "hybrid" parameters that combine the size of vertical features with the distance between them.

Most previous investigations on the efficacy of moisturizing products have relied on reduction of

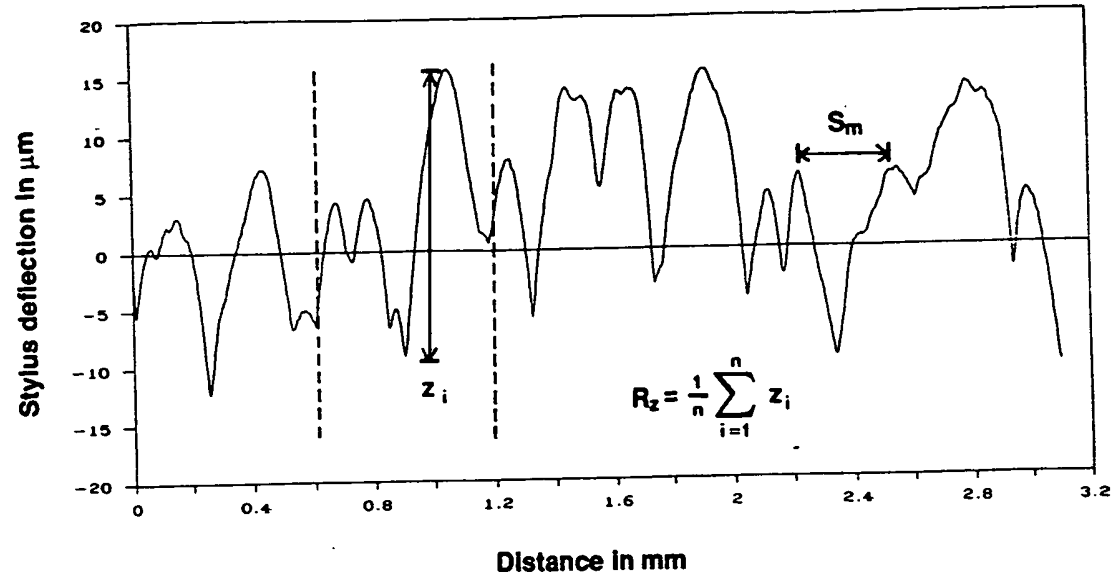

Figure 2: Sample of a profile obtained by scanning a skin replica surface with a diamond-tipped stylus, 20 µm in radius. S_m and R_z are two popular roughness parameters: S_m is the average peak spacing, R_z is the average amplitude of the profile.

the profile data to only amplitude parameters, as disclosed by the finding of a reduced number of peaks and the shift in the distribution to peaks of larger sizes.[16] At least one study has shown that application of a hydrating agent affects the horizontal spacing of peaks as well, so the average spacing between peaks decreases during the first two hours after moisturization, then returns to original values over the next six hours.[19]

This group of investigators also introduced a new horizontal parameter, the average width of the plateaus between major furrows, and described a reduction in plateau-size in the first two hours and a subsequent increase, but the measurement variance was lower than for the peak-spacing distribution.

Interestingly, the same article reports an increase in both the peak-spacing and the plateau-width with increasing age, just as with increasing skin dryness.

Roughness parameters

Actual hands-on analysis of profile lines can range from simple procedures that can be accomplished with a pen and a ruler, to more sophisticated mathematical approaches. In most cases the underlying concepts are straightforward, and the goals of the various types of analyses can be made apparent without resorting to the mathematical detail. Two of the more commonly used roughness parameters are:

R_a: The mean departure of the peaks and valleys of a profile from a reference line, the "mean line," and

R_z: the depth of the deepest valley plus the height of the highest peak, relative to the mean line, in one-fifth of the profile length, averaged over the entire profile.

A horizontal parameter commonly used is S_m: the average spacing of peaks. Each of these three parameters is calculated in straightforward fashion, and together they seem to provide a clear, even if biased, assessment of the skin profile. The calculation of R_z and S_m is illustrated in Figure 2.

There is a major flaw, however, in using such standard metallurgical parameters. The problem was first pointed out by Grove,[15] who compiled a series of pre-treatment and post-treatment skin measurements using parameters such as R_a, R_z and S_m in different scanning directions. It was found that the degree of change caused by a treatment ranged from 60 percent improvement to 23 percent worsening depending on the data selected. In his article, he concludes "... the marketer will understandably choose to use those parameters that yield the largest differences in favor of its product."

Our recent studies[21] have shown that even for *the same profile*, calculated metallurgical parameters vary substantially with the chosen length of measurement. For example, one can obtain any desired trend in R_z by varying the measured length. One approach used to avoid such inconsistencies is the use of "hybrid methods."

A hybrid-type analysis is one that combines both the amplitude and spacing of surface features. It requires simultaneous consideration of

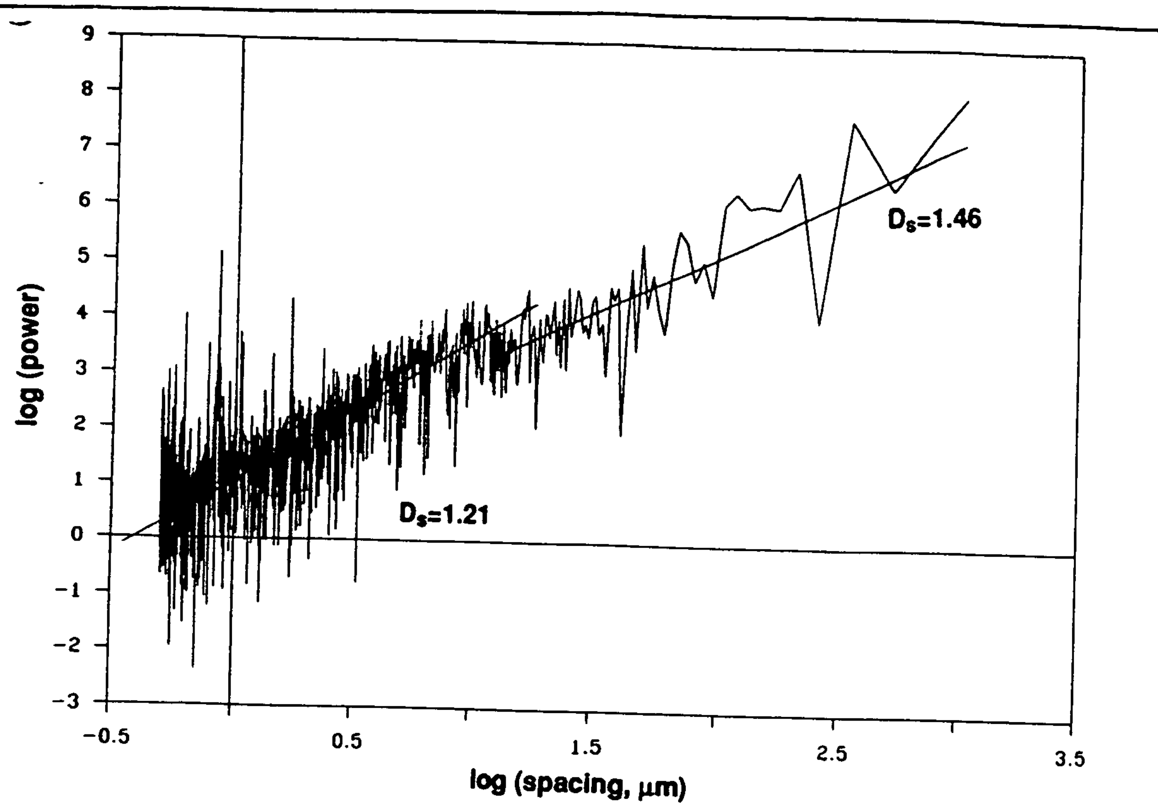

Figure 3: Fourier transform of a skin surface profile. Horizontal "spacing" refers to the wavelength; power indicates the vertical size of features of that length. The slope of this plot, D_s, is the "fractal dimension" of the surface, and indicates the roughness (see text).

both vertical and horizontal directions in the profile scan. This can be accomplished, for example, by regarding the multiple forms of undulations in the profile as discrete waveforms.

An ambitious application of wave theory to profilometry is the Fourier series representation of profile lines,[16] in which even the most irregular profile can be mathematically decomposed into a mixture of sinusoidal waveforms. Each waveform is characterized by its *wavelength*, which is a spacing parameter, and its corresponding *amplitude*, or height parameter. Examination of the relationship between amplitude and wavelength, across a wide wavelength spectrum, gives a sense for the distribution of surface features according to their lengths, i.e. horizontal dimensions.

Another approach

Fourier transformation retains all the information contained in the original profile (in a transformed form), and therefore is free of the problems plaguing the metallurgical roughness parameters. Its interpretation however, has been borrowed from analysis of *periodic* phenomena, such as waves or electrical signals. The rough surface of skin, as well as other rough surfaces, is *aperiodic*. This means that there is no single, dominating wave, and therefore the Fourier

power spectrum does not show outstanding peaks.

One method to overcome this difficulty is to use fractal geometry. The fractal approach to profilometry has been recently introduced by Zielinski.[21] Fractal analysis, like the Fourier transform, also includes all the information in the profile. Fractal geometry was introduced by B. Mandelbrot,[22] who used this name to describe a variety of mathematical structures that did not fit well to familiar (Euclidean) geometry.

The basic parameter used to characterize such structures is the "fractal dimension." Most people are familiar with the concept of dimensionality: a line is one dimensional ($D = 1$), a plane is two dimensional ($D = 2$), and a solid body three dimensional ($D = 3$). Fractal objects are such that their dimensionality is not a whole number. A fractal line is a convoluted, circuitous line with a dimension that therefore is higher than 1. The more convoluted it is, the more it fills the plane, and the higher its fractal dimension, which eventually can approach the value 2.

The fractal dimension can be used to characterize the roughness of a surface profile. A line with $D = 1$ is an ideally smooth line; the more rough the surface, the larger the value of D. The fractal dimension can have different values in different size ranges, as shown in Figure 3. There, the Fourier transform power spectrum was used to deduce the fractal dimension of a profile (there are a number of other methods too).

One can see that the "power," which is proportional to the square of the amplitude of surface features, does not show any distinct maxima. The *slope* of the line is, however, related to the original profile's fractal dimension. The value of $D_s = 1.21$ shows that *smaller* features, on the scale range from 0.5 to 5 um, are less "rough" than *larger* features, for which $D_s = 1.46$.

Fractal dimension, besides being a unique measurement of roughness, is also suited to distinguish features of different sizes, and hence the effects of treatment on specific morphological features.

Conclusions

Profilometry still is probably the best method for detecting and measuring morphological features of the skin surface as they relate to hydration phenomena. Whereas electrical measurements tend to be more sensitive to differences in skin hydration levels, the electrical properties also may reflect hydration in the deeper skin layers, not only surface properties. On the other hand, electrical measurements are easier to perform, and are in some respects more standardized than profilometry.

For example, whereas most investigators use the two-step replication technique, other techniques have been reported that either bypass skin replication altogether,[23] or rely only on stylus tracings of the negative impression.[3,24]

As already noted, there is some question whether the scaling typical of dry skin can be replicated satisfactorily. On the technical side, the type of stylus used, e.g. its weight and tip geometry, can influence the results. It is expected that stylus characteristics affect the way in which peaks and valleys of the surface are detected. The number and direction of measurements on the skin surface also must be considered, because body-surface markings tend to be both oriented and inhomogeneous, as well as site-dependent.

To obtain a fair sense of the surface contour, numerous parallel scans are required, but the number of scans varies greatly among published investigations. Moreover, the statistical criteria for acceptance of a given scan into the analysis also varies. We touched on the issue of selecting the most appropriate data reduction technique, and conclude that parametric interpretations of the skin surface profile are still a prime subject for scientific research and debate.

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